

# Delaware's Experience with Cyanobacteria in Freshwater Ponds

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## Introduction

With the increased emphasis on HABs in estuarine environments and their potential to impact natural resources and human health, the State DNREC in 2001 initiated a program to evaluate commercially available Microcystin kits and to measure Microcystin concentrations in select freshwater ponds. Although no environmental or human health impacts associated with blue-green algal blooms have been reported, Delaware does have a large number of private and public freshwater ponds which are accessible to humans, domestic animals, and wildlife.

A Standard Operating Procedure was developed for the EnviroLogix analysis tools in order to insure accurate repeatable results. The EnviroLogix kits are based on the presence of Microcystin LR in combination with Microcystin LA, Microcystin RR, Microcystin YR and Nodularin.

## Hypothesis

Since records, compiled by the State of Delaware Division of Fish and Wildlife, indicate that 14 out of 38 ponds have historically exhibited blue-green algae blooms, some of which included extensive masses of scum; it appeared possible that Microcystin concentrations could be elevated during bloom events. Since hepato-toxins produced by blue-green algae *Microcystis* and *Anabaena* had not been tested previously in Delaware freshwater ponds, it was necessary to collect data in order to demonstrate a need for routine monitoring of these surface waters.

## Methods

Surface water samples were collected from 6 freshwater ponds within the State which historically exhibited blue-green algal blooms. Samples were collected adjacent to the shoreline and in surface scum when present. Presence of scum anywhere in the pond on the day of collection was recorded. Ambient water samples were analyzed for the predominance of *Microcystis* and *Anabaena* via light microscopy (100X using a 0.1 Palmer Cell Counter) and for Microcystin concentrations using commercially available Microcystin analysis kits: EnviroLogix Microcystin Tube Kit ET022 (years 2002 & 2003), Strategic Diagnostic EnviroGard Microcystin Plate Kit (2002), and EnviroLogix Microcystin Plate Kit EP022 in conjunction with the Bio Tek  $\mu$ Quant Spectrophotometer Plate Reader Flx800 using KC4 software (2003). Intracellular Microcystin was released by freezing the water samples for a minimum of 24 hours but less than 1 month.

## Results

High concentrations of the organism *Microcystis* is NOT necessarily a good indicator of the level of Total Microcystin in water sample.

Two out of 88 samples from 9 ponds showed Dissolved Microcystin  $> 3$  ppb and 3 out of 88 showed concentrations  $> .5$  ppb but  $< 3$  ppb using the EnviroLogix Tube Kit. This occurrence was noted in 5 separate ponds all of which were sampled on September 24, 2002.

Five out of 18 samples analyzed with the EnviroGard Microcystin Plate Kit showed Total Microcystin  $> 1.00$  ppb, this is the provisional upper limit of Microcystin LR established by the World Health Organization for finished drinking water. Samples showing this exceedence (1.28 ppb to 3.28 ppb) were collected in late September (Sept. 26 and 30, 2002) at 4 separate ponds.

One out of 33 samples from 10 ponds showed Total Microcystin concentrations  $> 1$  ppb with the EnviroLogix Plate Kit, and 1 out of 33 samples showed Dissolved Microcystin  $> 0.5$  but  $< 3.0$  ppb. These results are from late August and mid-September respectively and from 2 separate ponds in 2003.

## Conclusions

Surface scum of freshwater ponds showed high concentrations of *Microcystis* 88% of the time, but had measurable levels of Microcystin ( $> 0.5$  ppb) only 17% of the time.

Total Microcystin measures both the Microcystin free in the water-column and that sequestered in the blue-green algae cells and as such its determination is recommended if Dissolved Microcystin concentrations approach 1 ppb.